

## CHROMOSOME NUMBER IN LIQUIDAMBAR

FRANK S. SANTAMOUR, JR.<sup>1</sup>

The genus *Liquidambar* belongs to the Hamamelidaceae, although most authorities consider this genus so distinct that they place *Liquidambar*, along with some other genera such as *Bucklandia* and *Disanthus*, in the sub-family Liquidambaroideae. Cytologically, all genera of Hamamelidaceae (sub-family Hamamelioideae) reported thus far have a basic chromosome number of  $x=12$  (Darlington and Wylie, 1955). *Liquidambar* is the only genus of the sub-family Liquidambaroideae that has been studied cytologically.

Anderson and Sax (1935) studied meiosis in *L. styraciflua* L. growing at the Arnold Arboretum in Jamaica Plain, Massachusetts. They concluded that *L. styraciflua* was a diploid ( $2n=30$ ) with a basic number of  $x=15$  chromosomes. They also noted marked meiotic irregularities and high pollen sterility in their material, and ascribed these irregularities to cultivation in an area north of the natural species range.

Pizzolongo (1958) reported that the diploid number of *L. styraciflua* (from shoot apices) and *L. orientalis* L. (from root tips) was  $2n=32$ , and concluded that the haploid (and basic) number was  $n=16$ . However, the only countable meiotic figure he found in *L. styraciflua* was an anaphase I, showing groups of 15 and 16 chromosomes. Meiotic irregularities and consequent pollen sterility apparently depended "upon some univalent chromosomes which do not respect the metaphasic congression and cause an unequal chromosome distribution among the pollen grains."

This study was conducted in Italy, where the trees, as in the Anderson and Sax work, were growing outside their native range.

<sup>1</sup>Research Geneticist, U. S. National Arboretum, Plant Science Research Division, Agricultural Research Service, U.S. Department of Agriculture, Washington, D.C. 20002.

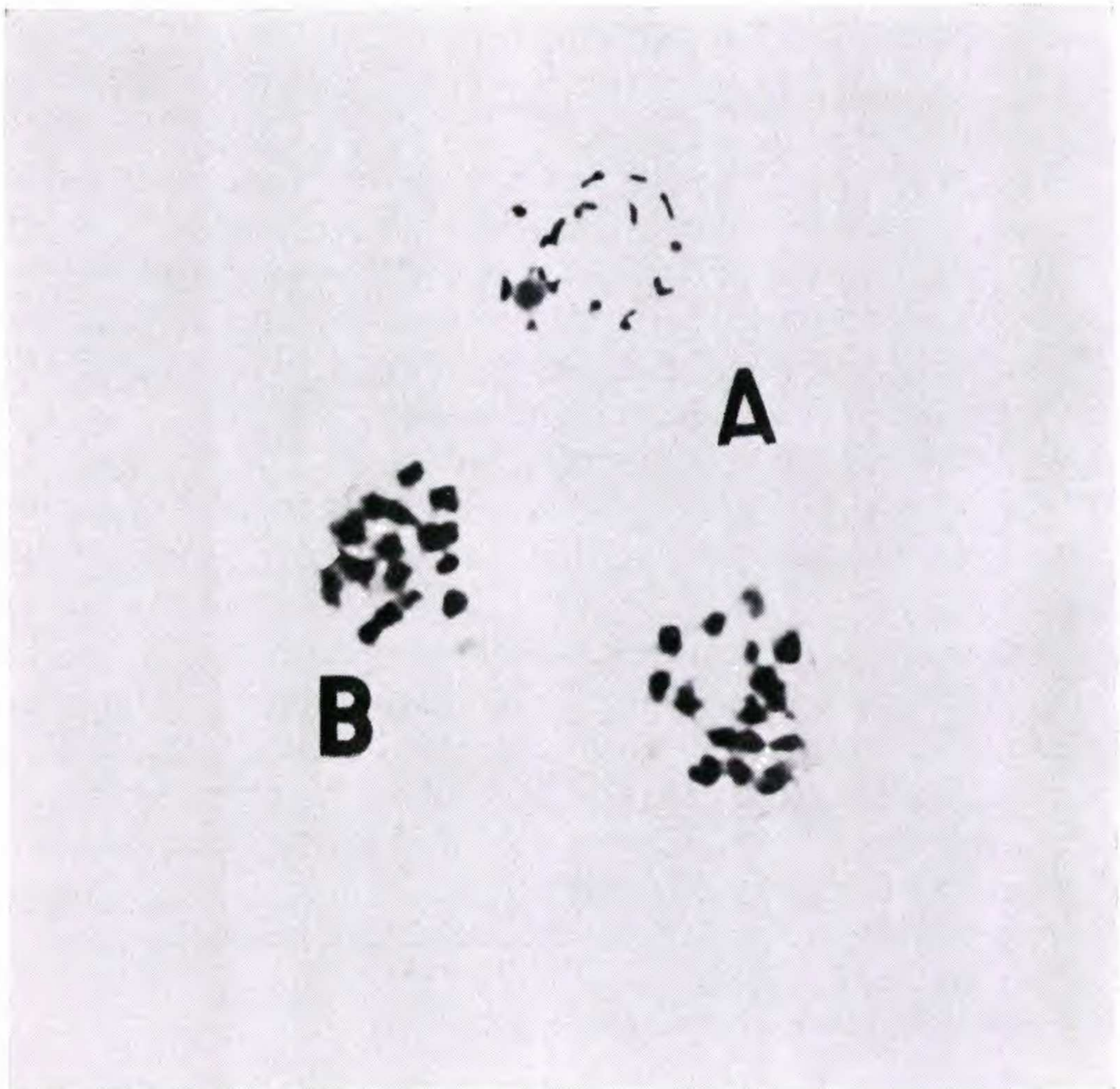


Figure 1. Meiotic stages in *Liquidambar styraciflua* L.;  
(a) Diakinesis with  $n=16$  (540  $\times$ )  
(b) Anaphase I with two groups of 16 chromosomes (1200  $\times$ ).

however, Pizzolongo did not believe that the species had two biotypes, with  $2n=30$  and  $2n=32$  chromosomes.

Work with *Liquidambar* in the project on "Cytogenetics, Breeding, and Evaluation of Shade Trees" at the U.S. National Arboretum has stressed interspecific hybridization. We have obtained hybrids between *L. styraciflua* and both Asiatic species, *L. orientalis* and *L. formosana* Hance. Thus, it was important to determine the number and meiotic behavior of the chromosomes of our parent trees, especially *L. styraciflua*, to decide if detailed cytological analysis of the progenies would be necessary.

In the spring of 1971, inflorescences of *L. styraciflua* were collected from 4 native trees on the grounds of the Arboretum in Washington, D. C. Two of these trees were used as parents in the breeding work. The flowers were fixed in 1:3 acetic-alcohol for 24 hours and stored in 80% ethanol. Meiosis in the pollen-mother-cell was studied using standard aceto-carminic squash techniques. Pollen was collected from the same trees and pollen abortion was determined from a random sample of 200 grains stained with aceto-carminic. Pollen size was based on measurements of 50 sound pollen grains from each tree. Mitosis in root tips of seedlings of *L. styraciflua* × *L. orientalis* and *L. styraciflua* × *L. formosana* parentage was also examined using aceto-carminic squash techniques.

Accurate chromosome counts at first metaphase of meiosis were, as noted by previous authors, impossible to achieve. The chromosomes at this stage are clumped together and no more than 10 distinct bivalents could be observed. Usually one, and frequently two, bivalents were found outside the metaphase grouping and exhibited precocious separation. Pizzolongo attributed the high pollen abortion he observed to this irregular chromosome behavior.

In my material it was not difficult to determine  $n=16$  chromosomes at diakinesis or late anaphase I. Many diads were counted with this number in both nuclei. At the 4-nucleate stage (anaphase II), nuclei with  $n=16$  chromosomes could also be determined, but in no case could all four nuclei be counted.

The major problem in documentation was obtaining figures in which all the chromosomes were in the same plane for photographic purposes. The picture of diakinesis (Fig. 1a) shows 16 chromosomes, with at least three and perhaps four chromosomes attached to the nucleolus. This configuration explains the multinucleolate microspores noted by Pizzolongo. At anaphase I (Fig. 1b), two groups of 16 chromosomes are shown.

Pollen abortion in the four trees studied ranged from 6% to 20% and averaged 11%. Pollen grain diameter averaged about 39 microns, and did not differ significantly among trees. Although abnormal meiotic behavior may explain the rather high pollen abortion sometimes found in this species, the degree of meiotic irregularities probably depends on individual tree characteristics.

Chromosome counts on root tips of hybrids of *L. styraciflua* with *L. orientalis* and *L. formosana* gave  $2n=32$ . These facts imply that all three species have the same chromosome number.

#### LITERATURE CITED

- ANDERSON, EDGAR & KARL SAX. 1935. Chromosome numbers in the Hamamelidaceae and their phylogenetic significance. *Jour. Arnold Arb.* 16: 210-215.
- DARLINGTON, C. D. & A. P. WYLIE. 1955. *Chromosome Atlas of Flowering Plants* (ed. 2) George Allen and Unwin Ltd., 519 pp.
- PIZZOLONGO, PAOLO. 1958. Ricerche cario-tassonomiche su alcune Hamamelidales. *Ann. Bot. (Roma)* 26: 1-18.